REMARKS

1. Support for the amendments and new claim

The amendments to claims 30 and 64, as well as new claim 66, are supported, for example, on page 69 lines 6-19 and page 69 line 21 to page 71 line 3, and thus the amendments do not constitute new matter. During a conference call with the examiner and her supervisor on June 30, 2004, it was agreed that the amended claim, with its recitation of image analysis techniques for cytoplasmic and membrane masking was supported in the specification as filed.

2. Rejection under 35 USC 112, second paragraph

The first paragraph of 35 USC §112 requires that the specification shall contain a written description of the invention. "To satisfy the written description requirement, a patent specification must describe the claimed invention in sufficient detail that one skilled in the art can reasonably conclude that the inventor had possession of the claimed invention." MPEP 2163, discussing *Vas-Cath Inc., V. Mahurkar,* 935 F.2d 1555, 1563 (CAFC 1991). "An applicant shows possession of the claimed invention by describing the claimed invention with all of its limitations using such descriptive means as words, structures, figures, diagrams, and formulas that fully set forth the claimed invention." MPEP 2163, discussing *Lockwood v. American Airlines, Inc.,* 107 F.3d at 1565, 1572 (CAFC 1997).

"The examiner has the initial burden of presenting evidence or reasoning to explain why persons skilled in the art would not recognize in the original disclosure a description of the invention defined by the claims." MPEP 2163 (II)(3)(b) citing *Wertheim*, 541 F2d at 263.

During a conference call with the examiner and her supervisor on June 30, 2004, it was agreed that amended claim 30 and its recitation of separate cytoplasmic and plasma membrane masking by use of the recited image analysis techniques was supported in the specification. For the sake of completeness, the Applicants will nonetheless respond to the rejection.

In the present case, the patent office has asserted that the amendment made in the office action response filed January 23, 2004 constituted new matter. Specifically, the patent office stated that:

> "Consideration of the support pointed to by Applicants regarding amending the claims to cite 'plasma' membrane instead of 'cell' membrane on pages 68-70 reveals that this amendment is NEW MATTER. No masking is cited on page 68. On page 69, lines 11-14, images of probes used to mark the 'plasma

membrane and cytoplasm' are used to mask the image... This supports a mask that is inclusive of both the plasma membrane and cytoplasm. No separate mask is cited such as now is present in the amended claims. On page 70, lines 15-18, similarly a masking of both plasma membrane and cytoplasm is again cited. No separate plasma membrane versus cytoplasm masking has written support."

The Applicants traverse these assertions. On page 69, lines 6-18, the application states:

> **Rho-RhoGDI complex translocation to the membrane.** In another embodiment, indicator cells are treated with test compounds and then fixed, washed, and permeabilized. The indicator cell **plasma membrane, cytoplasm,** and **nucleus** are all **labeled with distinctly colored markers** followed by immunolocalization of Rho protein (Self et al. (1995), *Methods in Enzymology* 256:3-10; Tanaka et al. (1995), *Methods in Enzymology* 256:41-49) with antibodies labeled with a fourth color. **Each of the four labels is imaged separately using the cell screening system,** and the images used to calculate the amount of inhibition or activation of translocation effected by the test compound. To do this calculation, the **images of the probes used to mark the plasma membrane and cytoplasm** are used to **mask the image of the immunological probe marking the location of intracellular Rho protein.** The integrated brightness per unit area under **each mask** is used to form a translocation quotient by dividing the **plasma membrane integrated brightness/area by the cytoplasmic integrated brightness/area.** By comparing the translocation quotient values from control and experimental wells, the percent translocation is calculated for each potential lead compound.

Thus, the different cell compartments (including cell cytoplasm and plasma membrane) are separately imaged, and the images of the markers for the cytoplasm and plasma membrane are used to create **separate** masks to determine the amount of intracellular Rho protein in the cytoplasm and the plasma membrane, as clearly evidenced by use of the term "each mask" when stating that "the integrated brightness per unit area under **each mask** is used to form a translocation quotient by dividing the **plasma membrane integrated brightness/area by the cytoplasmic integrated/brightness area.**" The patent office has asserted that the use of the phrase "images of the probes used to mark the plasma membrane **and** cytoplasm are used to mask the image..." means that only a single mask that is inclusive of both the plasma membrane and cytoplasm is made. This is an inaccurate reading of the phrase. Separate images of the cell cytoplasm and plasma membrane are made, and thus the images (plural because two separate images were made, one from a cytoplasm reporter and one from a plasma membrane reporter) are used to mark plasma membrane and cytoplasm (use of "and" to

indicate that the multiple images were used to make a plasma membrane mask **and** a cytoplasm mask). This interpretation is clearly supported by the subsequent statement that "the integrated brightness per unit area under **each mask** is used to form a translocation quotient by dividing the **plasma membrane** integrated brightness/are by the **cytoplasmic** integrated/brightness area." The use of the term "**each**" clearly teaches that there are separate cytoplasmic and plasma membrane masks. Similarly, if there was only a single mask, there would be no way to divide the plasma membrane integrated brightness/are by the cytoplasmic integrated/brightness area.

Thus, one of skill in the art would clearly understand that the specification does teach separate cytoplasm and plasma membrane masking, in contrast to the assertions made by the patent office.

A further example on page 69 line 20 to page 71 line 3 states:

> *β-Arrestin translocation to the plasma membrane upon G-protein receptor activation.*
>
> In another embodiment of a **cytoplasm to membrane translocation high-content screen**, the translocation of β-arrestin protein **from the cytoplasm to the plasma membrane** is measured in response to cell treatment. To measure the translocation, living indicator cells containing luminescent domain markers are treated with test compounds and the movement of the β-arrestin marker is measured in time and space using the cell screening system of the present invention. In a preferred embodiment, the indicator cells contain luminescent markers consisting of a green fluorescent protein β-arrestin (GFP-β-arrestin) protein chimera (Barak et al. (1997), *J. Biol. Chem.* 272:27497-27500; Daaka et al. (1998), *J. Biol. Chem.* 273:685-688) that is expressed by the indicator cells through the use of transient or stable cell transfection and **other reporters used to mark cytoplasmic and membrane domains**. When the indicator cells are in the resting state, the domain marker molecules partition predominately in the plasma membrane or in the cytoplasm. In the high-content screen, these markers are used to **delineate the cell cytoplasm and plasma membrane in distinct channels of fluorescence**. When the indicator cells are treated with a test compound, the dynamic redistribution of the GFP-β-arrestin is recorded as a series of images over a time scale ranging from 0.1 s to 10 h. In a preferred embodiment, the time scale is 1 h. Each image is analyzed by a method that **quantifies the movement of the GFP-β-arrestin protein chimera between the plasma membrane and the cytoplasm**. To do this calculation, the **images of the probes used to mark the plasma membrane and cytoplasm** are used to mask the image of the GFP-β-arrestin probe marking the location of intracellular GFP-β-arrestin protein. The integrated brightness per unit area under **each mask** is used to form a translocation quotient by **dividing the plasma membrane integrated brightness/area by the cytoplasmic integrated brightness/area**. By

> comparing the translocation quotient values from control and experimental wells, the percent translocation is calculated for each potential lead compound. The output of the high-content screen relates quantitative data describing the magnitude of the translocation within a large number of individual cells that have been treated with test compounds of interest.

Thus, the different cell compartments (including cell cytoplasm and plasma membrane) are separately imaged ("delineated"), and the images of the markers for the cytoplasm and plasma membrane are used to create **separate** masks to determine the amount of GFP-β-arrestin protein in the cytoplasm and the plasma membrane, as clearly evidenced by use of the term "each mask" when stating that "the integrated brightness per unit area under **each mask** is used to form a translocation quotient by dividing the **plasma membrane integrated brightness/area by the cytoplasmic integrated/brightness area.**" The patent office has asserted that the use of the phrase "images of the probes used to mark the plasma membrane **and** cytoplasm are used to mask the image..." means that only a single mask that is inclusive of both the plasma membrane and cytoplasm" is made. This is an inaccurate reading of the phrase. Separate images of the cell cytoplasm and plasma membrane are made, and thus the images (plural because two separate images were made, one from a cytoplasm reporter and one from a plasma membrane reporter) are used to mark plasma membrane and cytoplasm (use of "and" to indicate that the multiple images were used to make a plasma membrane mask and a cytoplasm mask). This interpretation is clearly supported by the subsequent statement that "the integrated brightness per unit area under **each mask** is used to form a translocation quotient by dividing the plasma membrane integrated brightness/are by the cytoplasmic integrated/brightness area." The use of the term "**each**" clearly teaches that there are separate cytoplasmic and plasma membrane masks. Similarly, if there was only a single mask, there would be no way to divide the plasma membrane integrated brightness/are by the cytoplasmic integrated/brightness area.

Thus, one of skill in the art would clearly understand that the specification does teach separate cytoplasm and plasma membrane masking, in contrast to the assertions made by the patent office.

Based on all of the above, the Applicants respectfully request reconsideration and withdrawal of this rejection.

2. Rejections under 35 USC 103

It is well established that the Patent Office bears the initial burden of establishing a *prima facie* case of obviousness, before any rejection under § 103 may be made. *In re Bell*, 991 F.2d 781 (Fed. Cir. 1993). Accordingly, "if the examiner does not produce a *prima facie* case, the applicant is under no obligation to submit evidence of non-obviousness." M.P.E.P. § 2142. Citing the Federal Circuit, the M.P.E.P. outlines three basic criteria that must be met to establish a *prima facie* case of obviousness, including the criterion that the prior art reference must teach or suggest all of the claim limitations.

During a conference call with the examiner and her supervisor on June 30, 2004, it was agreed that amended claim 30 was not obvious over the cited Cabib et al. reference. Nonetheless, for the sake of completeness, the Applicants will respond to the rejection.

A. The Patent Office has not established a *prima facie* case that Claim 30 is obvious over Cabib in light of *In re Venner*

Claims 30, and its dependent claims 44, 54, and 61-65, were rejected under 35 U.S.C. 103(a) under the assertion that they are unpatentable over Cabib et al. (US 5,784,162) in view of *In re Venner*, which is cited solely for the proposition that it is obvious to those of ordinary skill in the art at the time the invention was made to place analysis software on a machine readable medium for a manual activity. The Applicants respectfully traverse this assertion.

Presently pending claim 30 has been amended, and the cited Cabib reference neither teaches nor suggests at least the following limitations of currently pending claim 30:

> -creating a plasma membrane mask and a cell cytoplasm mask in the individual cells from the fluorescent signals from the plurality of fluorescent reporter molecules;
>
> -determining an intensity of the fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the plasma membrane mask and the cell cytoplasm mask in the individual cells in response to contacting the cells with a test stimulus;
>
> -determining a first translocation quotient between the cell cytoplasm and the plasma membrane for the cellular macromolecule of interest by calculating a ratio of the intensity of the fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the plasma membrane mask and the intensity of the fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the cell cytoplasm mask in the individual cells in response to contacting the cells at a first time point with a test stimulus;

-comparing the first translocation quotient to:

i) one or more second translocation quotients for the cellular macromolecule of interest between the cell cytoplasm and the plasma membrane, which are determined by calculating a ratio of an intensity of fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the plasma membrane mask and an intensity of fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the cell cytoplasm mask in the individual cells in response to contacting the cells with the test stimulus from at least a second time point; and/or

ii) one or more third translocation quotients for the cellular macromolecule of interest between the cell cytoplasm and the plasma membrane, which are determined by calculating a ratio of an intensity of fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the plasma membrane mask and an intensity of fluorescent signals from the fluorescent reporter molecules that report on the one or more cellular macromolecule of interest within the cell cytoplasm mask in the individual cells that have not been contacted with the test stimulus; and

g) determining the effect of the test stimulus on the distribution of the one or more cellular macromolecule of interest between the plasma membrane and the cell cytoplasm in the individual cells as a function of the first translocation quotient, the one or more second translocation quotients, and/or the one or more third translocation quotients.

Thus, the combination of cited references fails to teach all (or even many) of the limitations of currently pending claim 30, and thus does not meet the requirements for establishing a case of prima facie obviousness under MPEP § 706.02(j). Similarly, the dependent claims recite further limitations on claim 30, which are also not taught or suggested by the combination of the cited references. Therefore, the Applicants respectfully request reconsideration and withdrawal of this rejection.

B. The Patent Office has not established a *prima facie* case that Claims 30, 44, 54, and 61-65 are obvious over Harris et al. in light of *In re Venner*

Claims 30, and its dependent claims 44, 54, and 61-65, were rejected under 35 U.S.C. 103(a) under the assertion that they are unpatentable over Harris et al. (US 6,388,788) in view of *In re Venner*. The Applicants respectfully traverse this assertion.

Specifically, the patent office asserts that the priority date for the claims reciting "plasma membrane" is the actual filing date (11/27/00) of the present application, presumably

based on the patent office's assertion that there is a lack of written description of reciting separate masking of the cytoplasm and the plasma membrane. This issue has been addressed above, and the Applicants have clearly demonstrated that the specification as filed provides adequate written description for the currently pending claims.

The earliest priority date for Harris et al. is March 16, 1998, while the present application is a divisional of US application 09/031,271 filed February 27, 1998, which claims further priority to earlier applications. The current application is identical to the 09/031,271 application, and is thus at least entitled to the February 27, 1998 priority date of the 09/031,271 application. As a result, the Harris et al. reference is not appropriate prior art, and the applicants respectfully request reconsideration and withdrawal of this rejection.

Based on the foregoing, the Applicants believe that the application is ready for allowance. If the Examiner believes that a telephone or personal interview would expedite prosecution of the instant application, the Patent Office is invited to call the undersigned attorney at (312) 913-2106.

Respectfully Submitted,

Date: 9/13/04

By: _____

David Harper
Reg. No. 42,636
McDonnell, Boehnen Hulbert & Berghoff
300 South Wacker Drive
Chicago, IL 60606